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Confirming QTLs and Finding Additional Loci Conditioning Sheath Blight Resistance in Rice Using Recombinant Inbred Lines

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ABSTRACT

One method for confirming the existence of quantitative trait loci (QTLs) is to identify loci of similar location and effect in multiple populations and/or environments. The literature contains two prior publications reporting the location of QTLs affecting resistance to sheath blight (SB) disease in rice (*Oryza sativa* L.), but lack of agreement between QTLs in the studies left all 12 unconfirmed, limiting the potential of marker-assisted selection of this trait with worldwide importance. The earlier linkage analyses were imprecise due to heterozygosity, segregation, and limited plot size and replication. We evaluated a replicated set of pure-breeding recombinant inbred lines (RILs) to increase reliability of the quantitative disease data and in turn improve accuracy of the QTL mapping. The RILs were $F_{2:10}$ descendants from an early-generation 'Lemont' × 'Teqing' population wherein SB resistance QTLs (SB-QTLs) were first identified. The present study confirmed the location and effect of six SB-QTLs, confirmed the existence but not the specific location of another, and identified eight new loci. Three of the confirmed QTLs were also found to be independent from undesirable plant height and maturity effects. This research demonstrated the importance of using replicated phenotypic data for reliably establishing the identity and effect of putative QTLs for complex traits such as SB resistance. Further marker development to facilitate marker-assisted selection of these three SB-QTLs is warranted.

Sheath blight, caused by the fungus *Rhizoctonia solani* Kühn, is one of the major foliar diseases of rice worldwide that severely impairs both grain yield and quality (Ou, 1985). *R. solani* is a soilborne fungus with a broad host range that includes weed species and several cultivated crops. Fungicides capable of controlling the disease are available, but field scouting, proper timing of application, and aerial application of chemicals to flooded rice fields increase the production cost and limit the success of chemical control of SB disease. Although the existence of genes with large effect on SB resistance has been reported in some lines (Xie et al., 1990; Pan et al., 1999), no single gene conferring complete resistance to the fungus has yet been identified in rice. Thus, the wide variation seen among rice varieties for resistance to this disease (Khush, 1977; Groth and Nowick, 1992) is generally considered to be due to multiple resistance genes and modeled as a polygenic quantitative trait (Sha and Zhu, 1989; Li et al., 1995a).

Traditional breeding techniques, especially recurrent selection, have been used successfully to develop rice lines with reduced susceptibility to SB (Marchetti et al., 1995; Marchetti et al., 1996; McClung et al., 1997). These techniques are based on phenotypic evaluation of resistance in inoculated field plots. Disease response is not consistent, however, even within inoculated research plots. This inconsistency is experienced as differences between replications as well as in year-to-year and location-to-location differences. The development of SB is sensitive to surrounding humidity and temperature, which are themselves affected by plant density, tiller number, leaf angle and length, lodging, and water depth. As these plant traits and/or water depth vary within and between plots, disease severity also varies. The nongenetic macro- and micro-environmental factors affecting disease severity are so numerous and of strong impact that one generally observes a range of disease severity even within plots containing genetically pure materials, such as released cultivars. Thus, the disease index developed by Marchetti and Bollich (1991) was designed to account for differences in disease severity observed between plants within single plots.

On a larger scale, with southern U.S. coastal weather patterns, very late and sometimes very early maturing genotypes appear resistant when they have actually developed during a time when the environment was not optimal for fungal development. Sclerotia float on the surface of the water in flooded rice paddies. *Rhizoctonia* infection occurs at the water line then spreads up the plant. Thus, even small (<5 cm) differences in flood depth and/or mature height between plots and genotypes can affect disease ratings by impacting the distance between point of infection and the panicle. Due to high macro-and micro-environmental sensitivity, phenotypic evaluation of SB resistance requires replication and relatively large amounts of seed and is thus limited to later breeding generations.

In an effort to provide breeders with a more effective selection procedure, two research groups (Li et al., 1995a; Zou et al., 2000) have identified SB-QTLs based on cosegregation between markers and disease response as observed in specific mapping populations. Li et al. (1995a) observed disease response in $F_{2:3}$ progeny plots derived from a cross between 'Lemont' and 'Teqing'; Zou et al. (2000) studied clonal F_2 progeny from a cross between 'Jasmine 85' and Lemont. There was little agreement between the SB-QTLs reported by the two studies. Just one QTL reported by Zou et al. (2000) on chromosome 11 was co-located with a subthreshold log

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Abbreviations: LOD, log (base 10) of the odds ratio; QTLs, quantitative trait loci; RILs, recombinant inbred lines; SB, sheath blight; SBR, sheath blight response rating.

(base 10) of the odds ratio (LOD) peak reported by Li et al. (1995a) (shown in Fig. 1). Lemont was used as the susceptible parent in each of the two prior SB studies, and was the source of the resistance allele for this commonly identified locus on chromosome 11 (Zou et al., 2000).

Molecular geneticists and breeders are understandably hesitant to divert time and resources toward use of QTL markers for crop improvement until the existence and effect of those QTLs are confirmed. QTL confirmation may come from documented phenotypic change after marker-assisted selection (Romagosa et al., 1999; Kabelka et al., 2002), coincident shift in trait and allele expression when the QTL or marker–allele has been isolated in NILs (Swarup et al., 1999), or from independent identification of similarly located SB-QTLs in more than one population and/or environment (Terry et al., 2000; Bohn et al., 2001; Foolad et al., 2001; Li et al., 2001). Since the ability to identify marker–gene associations is limited by the reliability of the phenotypic data, it is possible that the lack of agreement between the two SB-QTL studies was caused by the quality of phenotypic data gathered and evaluated. Segregation within several of the $F_{2:3}$ plots was both expected and noted by Li et al. (1995a), such that the assignment of a single disease rating per plot was sometimes difficult during that study. Zou et al. (2000) eliminated plant-to-plant genetic variance by working with clonal F_2 plants but plot size was small, consisting of only eight transplanted plants. In the present study, we used a replicated set of pure-breeding recombinant inbred lines to increase the reliability of the quantitative disease ratings, which would in turn improve the accuracy of the QTL mapping results. Our objective was to confirm the location and effect of the previously reported SB-QTLs and identify additional SB resistance loci as a step toward our long-term goal of facilitating marker-assisted selection.

MATERIALS AND METHODS

Rice Study Population and Linkage Map

This project used a set of 300 RILs in the F_{10} and F_{11} generations derived from a cross between Lemont (Bollich et al., 1985), a U.S. cultivar susceptible to *R. solani*, and Teqing (Li et al., 1995a), a highly tolerant rice cultivar developed in China. These RILs were derived by single seed descent (Pinson et al., 1999) from the $F_{2:3}$ population which Li et al. (1995a) used

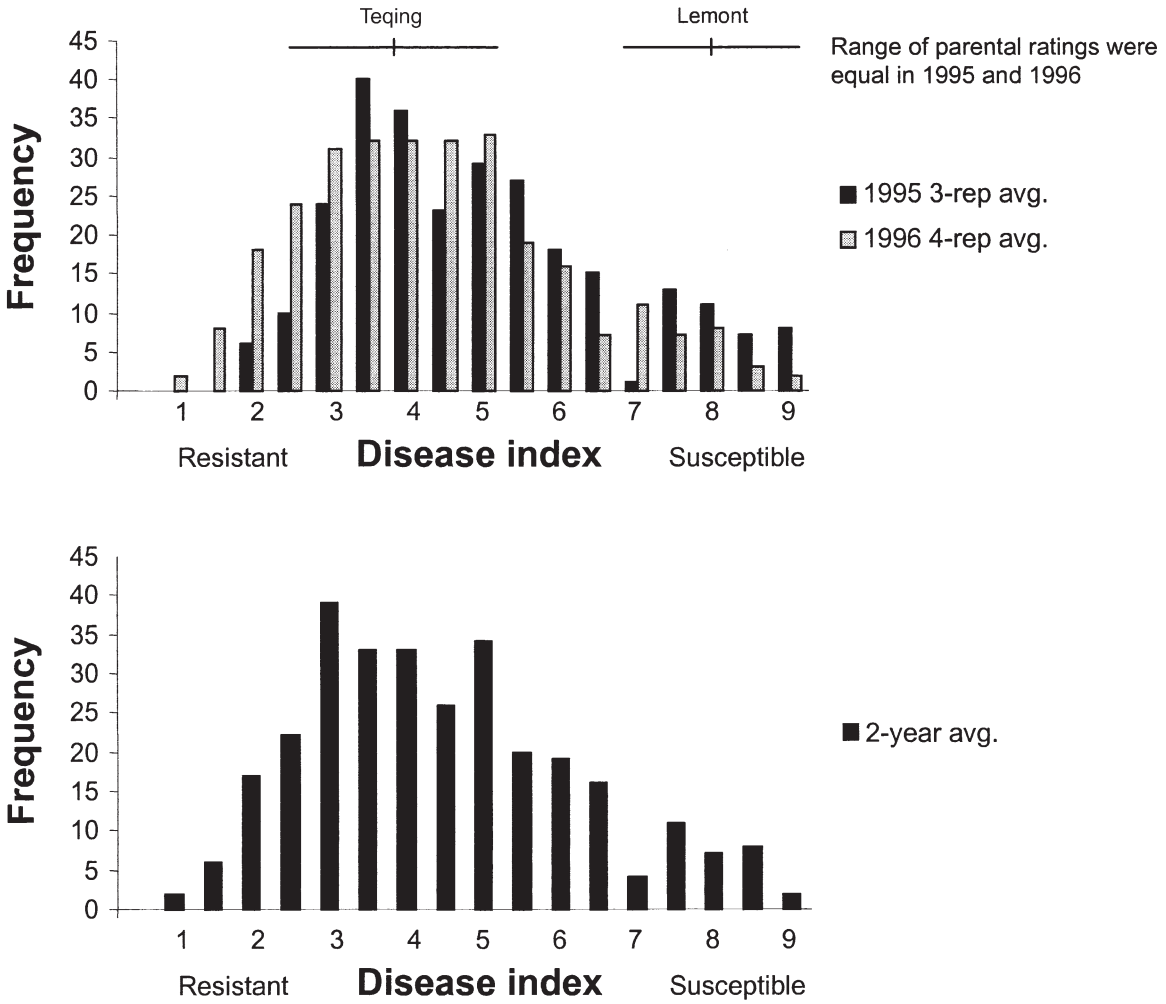

Fig. 1. The frequency distributions of sheath blight response ratings (SBR) of 300 recombinant inbred lines (RILs) from the cross 'Lemont' × 'Teqing' in a 2-year replicated field experiment exhibited continuous variation for SBR with skewing toward resistance both years.

to identify six SB-QTLs. The marker data and framework linkage map for these RILs were previously developed and used to locate major genes and QTLs for blast resistance (Tabien et al., 2000; Tabien et al., 2002). This map was formed using genotypic data from 284 F_8 generation RILs and was comprised of 173 RFLP-tagged loci plus two morphological markers distributed at an average of 10 centimorgans (cM; Kosambi, 1944).

Field Evaluation of Sheath Blight Resistance and Related Morphological Characters

Sheath blight resistance was evaluated in replicated field plots at the Texas A&M University System Agricultural Research and Extension Center in Beaumont, TX, following the procedure used by Li et al. (1995a). Seed of 300 RILs was drill-seeded into plots 2.4 m in length by three rows in width with 18 cm between rows and between plots. Three replications of F_{10} plants were observed in 1995 and four replications of F_{11} plants in 1996. Inoculum was prepared following Marchetti and Bollich (1991) and applied to plots approximately 60 d after planting. *R. solani* generally does not infect a plant severely until the host plant begins the grain filling process, when carbohydrates are remobilized upward to the developing kernels. As per Li et al. (1995a), the plots were rated weekly, then the disease score acquired approximately 30 d after heading for each plot was selected for analysis. A standard rating scale of 0 to 9 was used, where each unit of the scale approximates the proportion of aboveground plant length showing disease symptoms (Li et al., 1995a). A rating of 0 indicated no evidence of infection, 9 indicated that the plants were killed and collapsing, and 5 indicated that about 50% of the height of the plants above the water line was diseased.

Plant height and heading date were evaluated according to standard previously employed methods (Li et al., 1995b). Plant height was defined as the distance (cm) between the ground surface and the tip of the uppermost panicle and was measured in similarly grown but uninoculated plots replicated three times in nearby fields in the same years and generations as the SB plots. The number of days between planting and heading time was observed in both the inoculated and uninoculated plots.

Data Analysis

Analysis of variance of SB, height, and heading data indicated that replication effects were insignificant within years for each trait. Phenotypic data were therefore averaged over annual replications. Marker–trait linkage analyses were conducted using the two 1-yr averages plus a 2-yr mean phenotypic data set. The QTLs listed in Table 1 acquired a significant LOD score in one or more of the linkage analyses conducted for each trait.

Interval mapping was performed using MapMaker-QTL version 1.1 (Lander and Botstein, 1989) and followed the procedure of Tabien et al. (2002). A putative QTL was declared in a region having a LOD score ≥ 2.4 only when one or both markers flanking that LOD peak also attained a LOD ≥ 2.0. For each set of phenotypic data analyzed, the QTL having the largest LOD score was established, or fixed, as a cofactor in a multilocus genetic model within MapMaker-QTL, then the genome was rescanned to test for the presence of additional QTLs with smaller effects. The LOD threshold remained at 2.4 over background LOD for the rescanning process which was repeated until no new putative QTLs were detected. When a scan identified multiple peaks within a chro-

Table 1. Interval † and stepwise regression ‡ parameters for quantitative trait loci (QTLs) identified in a set of 300 recombinant inbred lines (RILs) derived from 'Lemont' and 'Teqing'. Sheath blight resistance scores were 2-yr means, compiling data from three replications in 1995 plus four replications in 1996.

Interval analysis†					Stepwise regression‡					
QTL	Marker nearest LOD peak	LOD§ value	% Variance§	Weight¶	Marker selected into model	Chromosome	Order into model	Partial R^2	F	$P > F$
qSB-1	RG532x	3.8	8	−0.55						
qSB-2	C624x	4.3	7	−0.67						
qSB-3-1	RG348x	12.1	18	−1.21	RG348x	3	1	0.257	24.86	<0.0001
		–#			RG450	3	4	0.052	6.85	0.0109
qSB-3-2	RZ474	4.8	10	−0.72						
qSB-4-1	RG1094e	3.0	5	−0.46						
qSB-4-2	RZ590x	4.6	7	−0.50						
qSB-5	Y1049	2.6	6	−0.55						
qSB-6-1	*C*	2.3	5	−0.30						
qSB-6-2	RZ508	4.3	7	−0.65						
		–#			CDO497	7	2	0.111	12.42	0.0007
qSB-7	C285	3.1	5	−0.38						
		–#			C424x	8	6	0.029	4.30	0.0419
qSB-8-1	G104††	0.0	0	0.82	G104	8	7	0.030	4.58	0.0360
qSB-8-2	R662††	0.6	1	0.53						
qSB-9	RZ404	3.8	6	−0.72	RZ404	9	3	0.054	6.48	0.0131
qSB-10	RG561	3.0	5	−0.53	RG561	10	5	0.040	5.61	0.0207
qSB-12	G1106	4.3	9	−0.62						
					G1468a	12	8	0.026	4.12	0.0466
Multilocus model	15 loci	28.5	41				8 loci	0.466		

† QTLs were declared based on interval analysis (LOD ≥ 2.4, MapMaker-QTL version 1.1). The number after "qSB-" indicates the chromosome on which that QTL resides.

‡ Stepwise regression considered all marker loci that had exhibited association in either interval analysis (LOD ≥ 2.0) or single factor ANOVA of any of the three traits measured here as well as all loci that had been associated with resistance to other diseases in early- or late-generation Lemont–Teqing gene-mapping populations (Li et al., 1995a; Li et al., 1995b; Li et al., 1999; Tabien et al., 2000; Tabien et al., 2002).

§ LOD scores and % variance explained are from interval analysis univariate models (no fixed QTLs).

¶ Weights and QTL locations are from multilocus interval analysis models. Estimated QTL weights are equivalent to additive effects and are expressed here in terms of the estimated change in SB rating expected from introgression of the Teqing allele into Lemont genome. A negative weight indicates that Teqing is the origin of the resistance allele.

When the nearby loci with higher LOD scores were first fixed in the genetic models; RG450, CDO497, and C424x did not increase LOD scores by 2.0, the threshold for declaring two linked QTLs.

†† qSB-8-1 and qSB-8-2 acquired LOD > 2.4 in interval analysis after fixing three loci, namely qSB-3-1, qSB-3-2, and qSB-9.

mosomal region, the peak with the larger LOD score was "fixed" and the genome was rescanned. Two loci were declared in that region if the LOD score for the 2-gene model exceeded the LOD score of the highest 1-gene model by 2.0 or more. MapMaker-QTL 1.1 considers a maximum of seven QTLs in any single model. The weight of each QTL was calculated from an interval analysis model that simultaneously included the six most definitive (largest LOD score) QTLs plus the QTL in question. Absolute values of QTL weights are equivalent to additive effect estimates and are presented here in terms of the magnitude of change in SB rating one would expect from the substitution of the Teqing allele into Lemont. Thus, a negative weight indicates that Teqing is the donor parent of the resistance allele.

Single factor ANOVA was performed using SAS PROC GLM (SAS Institute, 2000) with a F-test probability level set conservatively at 0.002 to minimize the occurrence of Type 1 error. Stepwise regressions were conducted in SAS to further evaluate the marker–trait linkages identified through interval analysis and single factor ANOVA. Independent variables considered in the stepwise regressions were the same for the analysis of each of the three phenotypic traits and included all the marker loci that (i) had exhibited a LOD peak $>$ 2.0 in the single and multilocus models analyzed by interval analyses for any of the three traits, (ii) had been significantly associated with one or more of the traits by single factor ANOVA, or (iii) was located in a chromosomal region previously reported to contain a SB-QTL (Li et al., 1995a; Zou et al., 2000). The significance threshold for the stepwise regression analyses was set at $P \geq 0.05$.

RESULTS AND DISCUSSION

Phenotypic and Genetic Variation for Response to *Rhizoctonia solani*

The Teqing and Lemont control data were quite similar over the two years. The SB response ratings (SBRs) of the individual Teqing plots ranged from 2 to 6 in both 1995 and 1996; Lemont plot ratings ranged from 7 to 9. Teqing's average ($\pm$ the standard deviation) in 1995 was 3.7 $\pm$ 1.1 and was 3.6 $\pm$ 1.0 in 1996. Lemont averaged 8.4 $\pm$ 0.8 in 1995 and 8.0 $\pm$ 1.0 in 1996. Individual RIL plot ratings ranged from 1 to 9 in both years and calculated 2-yr averages ranged from 1.25 to 8.75 (Fig. 1). Four RILs, namely LQ:163, LQ:200a, LQ:204a, and LQ:207, had yearly average ratings $\leq$ 2.0 in both years, suggesting they might be more resistant than Teqing. Each of these seemingly resistant RILs flowered 10 to 20 d later than Teqing, and the cooler night temperatures during this later maturation period (data not shown) may have contributed to this apparent resistance by slowing pathogen growth. These lines are also 10 to 25 cm taller than Teqing which could also have contributed to their apparent resistance.

An ANOVA indicated that differences between years for SBR ($R^2 = 0.009$) was small compared to the genetic differences between RILs ($R^2 = 0.47$). Even so, linkage analyses were conducted using SBR data averaged over individual years as well as using 2-yr averages.

Confirmed and Newly Identified QTLs Conditioning Sheath Blight Resistance

Fifteen SB-QTLs attained a significant LOD score during interval analysis of the RIL data (Table 1, Fig. 2). Teqing provided the resistance alleles for all loci except the two QTLs located on chromosome 8. Six of the presently identified SB-QTLs—referred to as qSB-2, qSB-3-1, qSB-3-2, qSB-4-2, qSB-8-1, and qSB-9 in Table 1 and Fig. 2—overlapped in genomic location with previously reported putative SB-QTLs (Li et al., 1995a; Zou et al., 2000). Whether the resistance allele originated from the U.S. cultivar Lemont or from the foreign indica variety at these six loci was also consistent with the previous QTL reports. In further agreement with the Li et al. (1995a) study which was based on an earlier generation of the Lemont and Teqing RILs reported here, qSB-3-1 displayed the highest LOD score and the largest additive effect among the QTLs identified in Lemont and Teqing. With independent identification and mapping location of these six SB-QTLs in multiple populations and environments, qSB-2, qSB-3-1, qSB-3-2, qSB-4-2, qSB-8-1, and qSB-9 can now be considered "confirmed" for existence and genomic location. Additionally, a QTL was presently detected on chromosome 7 near to, but not identically located with a QTL previously reported by Zou et al. (2000). The molecular map used in the present study contains gaps on chromosome 7 that prevent precise mapping of genetic factors in this genomic region. Thus, the existence of a SB-QTL on chromosome 7 can be considered confirmed but not its map location.

Among the 12 putative QTLs reported previously (Li et al., 1995a; Zou et al., 2000), only four (two on chromosome 9 and one each on chromosomes 11 and 12, Fig. 2) were not well supported by the QTLs identified via interval analysis of RIL data in the present study. Among the previously reported QTLs that were not confirmed by the present study was the location on chromosome 11 putatively identified in both of the earlier reports. Zou et al. (2000) reported a QTL with the resistance allele coming from the Lemont parent located on chromosome 11 where Li et al. (1995a) had earlier reported a LOD peak lower than their criteria for declaring a QTL. Single-factor ANOVA of the present RIL data also detected association between SB ratings and markers in this chromosomal region. However, interval analysis of the same phenotype and marker data sets did not support a QTL on chromosome 11.

Of the five previously reported Teqing and Lemont QTLs (Li et al., 1995a), two were not confirmed with the present analysis of Lemont and Teqing RILs. These loci, one located on chromosome 9 and the other on chromosome 12, produced the lowest LOD scores among the five previously reported QTLs. These loci were also reported to exhibit high dominance effect but relatively low additive effect. The lack of agreement between the earlier and the present study based on Lemont and Teqing progeny may be explained by the inbreeding and near-elimination of heterozygosity within the presently studied RILs.

The previously reported lack of agreement between the SB-QTLs identified by Li et al. (1995a) and Zou et al. (2000) suggested that their respective resistant lines, Teqing and Jasmine 85, had no SB-QTLs in common. After using replicated observation of RILs to map Teqing genes in better detail, we now see that Teqing has

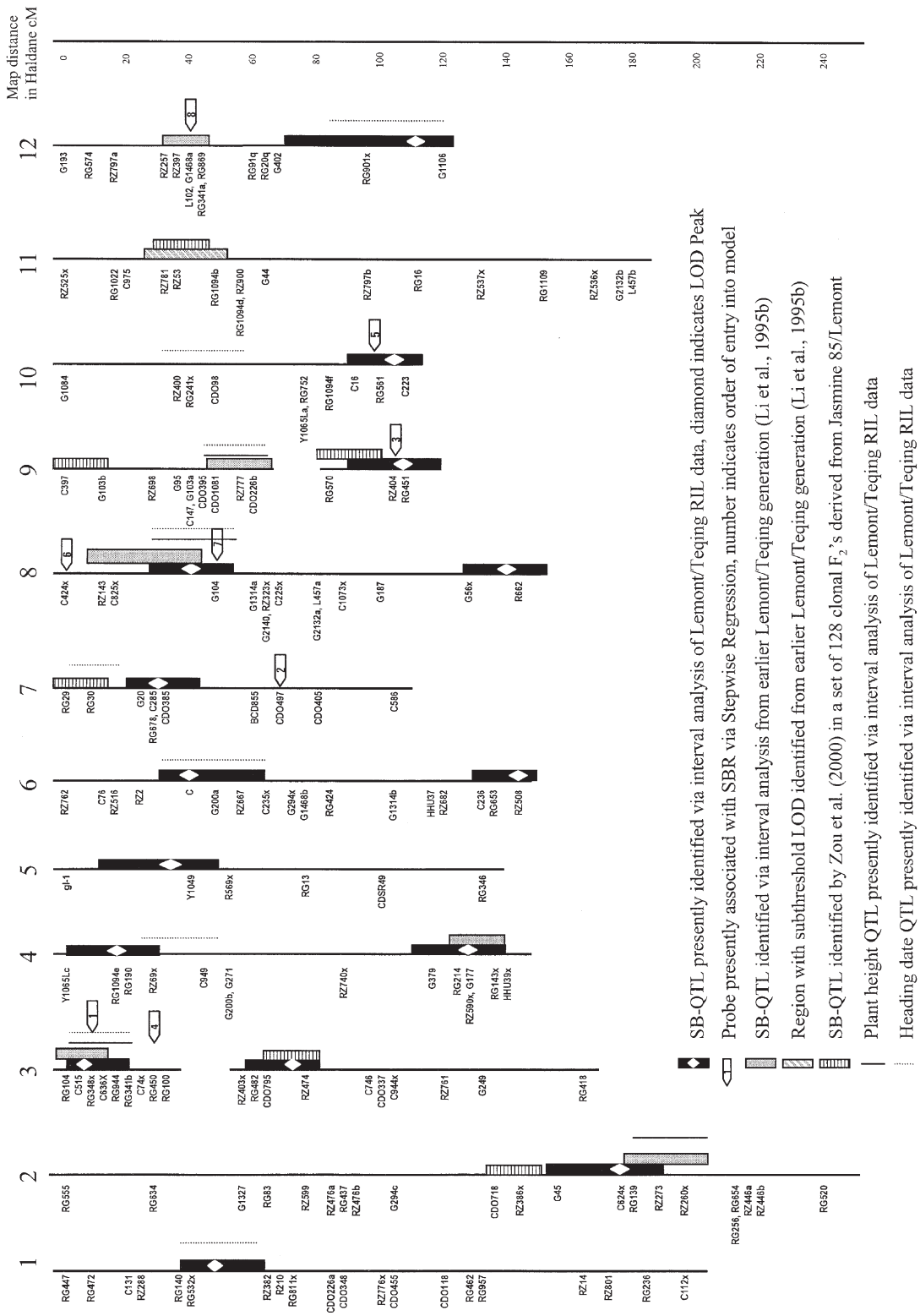

Fig. 2. Estimated location of rice quantitative trait loci (QTLs) affecting sheath blight (SB) resistance, plant height, and heading date identified from present analysis of recombinant inbred lines (RILs) derived from 'Lemont' and 'Teqing' and comparatively mapped with SB-QTLs from two prior studies involving F_2 and/or F_3 progeny (Li et al., 1995a; Zou et al., 2000). The shaded boxes cover the region where the QTLs are most likely located based on where the likelihood score was within 1 LOD of its maximal value.

SB-QTLs similarly located with four of the six Jasmine 85 loci reported by Zou et al. (2000) (Fig. 2). The present study also identified eight additional QTLs in chromosomal regions not previously reported to contain SB-QTLs; namely qSB-1, qSB-4-1, qSB-5, qSB-6-1, qSB-6-2, qSB-8-2, qSB-10, and qSB-12.

Stepwise regression was applied to further evaluate the markers and loci found by interval analysis and

single-factor ANOVA to be associated with SB resistance. Six of the eight markers selected by stepwise regression were located at or near four of the 15 QTLs presently identified via interval analysis (Table 1, Fig. 2), including three of the QTLs now considered confirmed via independent analysis. Stepwise regression did not clarify the location of a putative QTL on chromosome 7 as was hoped but identified instead a region even farther away from the originally reported SB-QTL than the QTL identified via interval analysis (Fig. 2). The marker selected by stepwise regression on chromosome 12, G1468a, was not located near qSB-12, the QTL identified from interval analysis of the same RIL data, but was instead similarly located with a Teqing resistance allele reported earlier by Li et al. (1995a). Interval analysis combines information from neighboring markers to estimate the location of QTLs, while ANOVA and regression analyses consider each marker individually and without chromosomal or linkage context. Therefore, interval analysis is generally considered to provide a better estimate of QTL location than ANOVA and regression analyses. In the present case, however, our ability to accurately determine the location of qSB-12 is limited by large gaps between markers contained in this region. These gaps in molecular coverage also limit our ability to accurately evaluate the existence of two versus one QTL in this region. It may be that two SB-QTLs reside on chromosome 12 with one being overshadowed by the statistically detected effects of the other locus in both the earlier and the present study.

Association between Sheath Blight QTLs and Plant Height and Heading Date

As was noted in the introduction section, tall plant height can bias SB disease severity ratings downward (toward resistance). Downward bias can also result from very early or very late maturity that allows plants to develop when the environment is less conducive to disease development. The fact that tall genotypes also tend to mature later further increases the probability that loci declared as "SB-QTLs" are indirectly affecting disease response through a more direct effect on plant height and/or maturity. Unfortunately, neither tall height nor late maturity are desired in modern U.S. rice cultivars due to lodging tendencies and other economic reasons. The SB-QTLs that result from direct effect on height and maturity are not as useful to breeders as the SB-QTLs disassociated with these other plant characters. Thus, as in the two prior SB-QTL publications, we evaluated association between the SB-QTLs with plant height and maturity. Because Teqing and Lemont are both semidwarf varieties containing the sd_1 semidwarf gene (Li et al., 1995b), all of the plant height genes in this Lemont–Teqing RIL population are expected to be "modifier genes" rather than major genes, with less individual effect than what breeders have come to expect from experience with the sd_1 gene. In the context of genes with small individual effect, estimates of additive gene effect will be more meaningful to rice breeders and geneticists than the portions of total variance explained by each resistance, height, or heading date QTL.

The two SB-QTLs presently identified as having strongest genetic weight (Table 1), equivalent to additive effect among the RILs, were qSB-3-1 and qSB-8-1. These SB-QTLs also had the highest LOD scores and relatively high additive effect in the earlier study on Lemont and Teqing SB-QTLs (Li et al., 1995a). In further agreement with that prior study, the resistance alleles at each of these loci were associated with both increased plant height and heading time with Teqing being the source of the resistant–tall–late allele for qSB-3-1 and Lemont being the source of the resistant–tall–late allele for qSB-8-1. Six of the 15 presently identified SB-QTLs had essentially identical LOD peaks for height and/or heading, including the already mentioned qSB-3-1 and qSB-8-1, plus qSB-1, qSB-2, qSB-6-1, and qSB-12 (Fig. 2). In two additional cases, one on chromosome 4 and another on chromosome 7 (Fig. 2), SB resistance appeared linked to, but not identically located with, alleles for late heading date. The seven remaining SB-QTLs (qSB-3-2, qSB-4-2, qSB-5, qSB-6-2, qSB-8-2, qSB-9, and qSB-10) appear to be independent of plant height and heading time.

Association between Sheath Blight QTLs and Loci Conferring Resistance to Other Rice Diseases

Clustering of disease resistance genes into resistance gene blocks, first noted in maize (Sudapak et al., 1993) and lettuce (Witsenboer et al., 1995), has been noted previously in rice studies (Song et al., 1997; Tabien et al., 2002). This report of rice SB-QTLs adds to this body of knowledge. Rice loci conferring resistance to bacterial leaf blight disease [causal organism *Xanthomonas oryzae* pv. *oryzae* (Ishiyama) Swings et al.] and rice blast (causal organism *Pyricularia grisea* Sacc.) have also been studied in the Lemont and Teqing RILs (Li et al., 1999; Tabien et al., 2002). Nine of the fifteen SB-QTLs reported here (qSB-3-1, qSB-3-2, qSB-4-1, qSB-4-2, qSB-6-2, qSB-7, qSB-8-1, qSB-9, and qSB-10) appear closely linked to loci affecting two or all three of the diseases studied in this same set of RILs. A blast resistance QTL was also reported on chromosome 12 near RG869 (Tabien et al., 2002). This region was not a SB-QTL according to interval analysis but was identified by stepwise regression as associated with SB resistance, lending supportive evidence to the existence of a disease resistance gene in this region. Loci associated with resistance to multiple diseases are of particular interest to pathologists and geneticists as well as breeders as they may provide clues to plant mechanisms that decrease the virulence of or increase the plant's ability to resist many microbial pests.

Impact on Future Breeding and Genetics Studies

While this study provides independent confirmation of association between seven chromosomal regions with SB resistance, this study also verifies that the resistance alleles at four of these loci are also associated with undesirably tall plant height and/or late heading time.

The high number of genes affecting SB resistance, plant height, and heading time segregating within the study population prevent the distinction between pleiotropy and close linkage, but both may cause difficulty for breeders. The remaining three confirmed resistance loci, qSB-3-2, qSB-4-2, and qSB-9, are of particular interest then, because their independence from undesirable height and maturity has also been confirmed. In terms of additive gene effect, qSB-3-2 and qSB-9 are relatively high, with each Teqing allele associated with a 0.72 reduction in SB score (weight = −0.72, Table 1). Substituting both Lemont alleles at one of these loci with resistant alleles from Teqing would reduce disease development by approximately 1.4 rating units, from an expected rating of 8 for Lemont down to 6.6 for the gene substitution line. Marchetti and Bollich (1991) reported the relationship between SB score and yield loss to be calculated as "percent yield loss = −1.8 + 5.1(SB score)". Gene substitution for one of the SB-QTLs would reduce anticipated yield losses from 40 to 32%; gene substitution at both qSB-3-2 and qSB-9 would further reduce yield losses to 25%. Adding to breeder interest in qSB-3-2 and qSB-9 are the fact that these loci are favorably linked with loci conferring resistance to other diseases such as blast (Tabien et al., 2002) and bacterial leaf blight (Li et al., 1999).

The RFLP markers identified here as associated with SB-QTLs would not be cost effective for evaluating relatively large numbers of breeding progeny. However, one can use the marker associations reported here and by earlier studies to identify candidate PCR-based markers, which are more economical, from the publicly available microsatellite and sequence databases including www.gramene.org/microsat/, www.ncbi.nlm.nih.gov/blast/Genome/PlantBlast.shtml?2, and www.tigr.org/tdb/e2k1/osa1/blastsearch.shtml. Linkage of the candidate markers to the confirmed QTLs, whether resistance originates from Lemont or Teqing, could be accomplished using the present set of Lemont and Teqing RILs. However, the high background variation caused by the segregation of many resistance, height, and heading genes within the RILs limits the ability to finely map any single SB-QTL. Because Teqing has been incorporated into several U.S. rice breeding populations, advanced breeding lines having introgressions from Teqing and already selected for desirable plant height and maturity may prove more immediately useful for fine mapping of SB resistance alleles originating from Teqing.

We will be employing this strategy to more finely map the SB-QTLs and develop molecular gene-tags that can be used to assist breeders in incorporating the confirmed SB-QTLs from Teqing into improved rice cultivars adapted to U.S. commercial rice production.

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